Appl. No. 10/584877 Reply to Office Action dated 1/26/09

612-455-3801

JUL 24 2009

Amendments to the Claims:

This Listing of Claims will replace all prior versions and listings of claims in the application. No new matter has been added.

Listing of Claims:

1. (Currently Amended) A method for the treatment or prevention of autoimmune arthritis comprising administering to a patient an effective amount of a coumestans compound of formula I or pharmaceutically acceptable salts thereof, or an extract containing the cournestans compound of formula I or pharmaceutically acceptable salts thereof

wherein

R₁ represents H, OH, or methoxyl;

R₂ represents H, OH, or C₁-C₈ alkyl;

R₃ and R₄ are each independently selected from the group consisting of H, halogen, OH, and methoxyl.

- 2. (Previously Presented) The method of claim 1, wherein the pharmaceutically acceptable salt of the compound of formula I are formed with the acids selected from the group consisting of chlorhydric acid, hydrobromic acid, sulfuric acid, citric acid, tartaric acid, phosphoric acid, lactic acid, pyruvic acid, acetic acid, succinic acid, oxalic acid, fumaric acid, maleic acid, ketosuccinic acid, methane-sulfonic acid, ethyl-sulfonic acid. benzene sulfonic acid, and isethionic acid.
- 3. (Previously Presented) The method of claim 1, wherein the compound of formula I or the extract is extracted from the plant Compositae.

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- 4. (Previously Presented) The method of claim 3, wherein the plant Compositae is selected from Eclipta prostrata Linn, Wedelia chinensis, and Eclipta alba.
- 5. (Previously Presented) The method of claim 1, wherein said compound of formula I is wedelolactone as shown in formula II:

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- 6. (Previously Presented) The method of claim 1, wherein the arthritis is selected from the group consisting of rheumatic arthritis, rheumatoid arthritis, and osteoarthritis.
- 7. (Previously Presented) The method of claim 1, wherein the compound of formula I is produced by an extraction method comprising the steps of:
- (a) extracting the fruits, leaves, or branches of the Compositae plant with 95±3% ethanol, thereby producing ethanol extract:
- (b) dissolving the above ethanol extract in 5-300 volumes of H₂O at 50-80°C, filtering to remove the precipitates and collecting the H₂O phase;
- (c) extracting the H₂O phase in step (b) with acetic ester and collecting the acetic ester phase;
- (d) concentrating and drying the acetic ester phase in step (c) to produce the precipitates;
- (e) eluting the precipitates in (d) on a silica gel column with a 5:1 to 1:2 gradient of petroleum ether/acetone mixture and collecting the fraction eluted with 1:1 petroleum ether/acetone;
 - (f) concentrating the fraction eluted in step (e) to produce concentrated residue;
- (g) eluting the concentrated residue in step (f) on a silica gel column with a 5:1 to 1:2 gradient of dichloromethane/acetone mixture and collecting the fraction eluted with 3:1 dichloromethane/acetone;

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- (h) eluting the fraction eluted in step (g) on a silica gel column with a 20:10:1 to 5:10:1 gradient of toluene-acetone-formate mixture and collecting the fraction eluted with 10:10:1 toluene-acetone-formate;
- (i) eluting the fraction eluted in step (h) on a silica gel column with a 30:1 to 1:1 gradient of dichloromethane/methanol mixture and collecting the fraction eluted with 20:1 dichloromethane/methanol; and
- (j) recrystallizing the fraction eluted in step (i) with ethanol, thereby producing the coumestans compounds of formula I as precipitates.
- 8. (Original and Withdrawn) A method for producing coumestans, comprising the steps of:
- (a) extracting the fruits, leaves, or branches of the *Compositae* plant with 95±3% ethanol, thereby producing ethanol extract;
- (b) dissolving the above ethanol extract in 5-300 volumes of H₂O at 50-80°C, filtering to remove the precipitates and collecting the H₂O phase;
- (c) extracting the H₂O phase in step (b) with acetic ester and collecting the acetic ester phase;
- (d) concentrating and drying the acetic ester phase in step (c) to produce the precipitates;
- (e) eluting the precipitates in (d) on a silica gel column with a 5:1 to 1:2 gradient of petroleum ether/acetone mixture and collecting the fraction eluted with 1:1 petroleum ether/acetone;
 - (f) concentrating the fraction eluted in step (e) to produce concentrated residue;
- (g) eluting the concentrated residue in step (f) on a silica gel column with a 5:1 to 1:2 gradient of dichloromethane/acetone mixture and collecting the fraction eluted with 3:1 dichloromethane/acetone;
- (h) eluting the fraction eluted in step (g) on a silica gel column with a 20:10:1 to 5:10:1 gradient of toluene-acetone-formate mixture and collecting the fraction eluted with 10:10:1 toluene-acetone-formate;

以是这个时间,这个时间是这个时间,这个时间,也是这个时间,我们是这种的时间,我们就是这种时间,也是是这种的时间,也是这种的,也是这种,我们也是这种的,我们就是这

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(i) eluting the fraction eluted in step (h) on a silica gel column with a 30:1 to 1:1 gradient of dichloromethane/methanol mixture and collecting the fraction eluted with 20:1 dichloromethane/methanol; and

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(j) recrystallizing the fraction eluted in step (i) with ethanol, thereby producing the coumestans compounds of formula I as precipitates

$$R_2$$
O R_3 R_4

wherein,

R₁ represents H, OH, or methoxyl;

R₂ represents H, OH, or C₁-C₈ alkyl;

R₃ and R₄ are each independently selected from the group consisting of H, halogen, OH, and methoxyl.

9. (Original and Withdrawn) A dietary supplement comprising 0.05-50wt% of the compound of formual I, or the pharmaceutically acceptable salts thereof:

R₁ represents H, OH, or methoxyl;

R₂ represents H, OH, or C₁-C₈ alkyl;

 R_3 and R_4 are each independently selected from the group consisting of H, halogen, OH, and methoxyl;

and a bromatologically acceptable carrier.

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- 10. (Original and Withdrawn) A pharmaceutical composition for treating arthritis, comprising:
- (a) 0.05-90wt% compound of formula I or pharmaceutically acceptable salts thereof, or extracts containing the coumestans compound of formula I or pharmaceutically acceptable salts thereof, as the main active ingredient,

R₁ represents H, OH, or methoxyl; and

R₂ represents H, OH, or C₁-C₈ alkyl;

R₃ and R₄ are each independently selected from the group consisting of H, halogen, OH, and methoxyl; and

- (b) one or more active ingredients selected from the group consisting of: accemetacin, diclofenac, ibuprofen, indomethacin, meloxicam, ketoprofen, sulindac, auranofin, naproxen, nabumetone, piroxicam, mecolfenamic acid, chlofenamic acid, mefenamic acid, pirprofen, fenbufen, tolmetin, flufenamide acid, methocarbamol, nimesulide, celecoxib, rofecoxib, aceclofenac methotrexate, gold salts, salazosulfadimidine, penicillamine, chloroquine, tripterygium wilfordii, ciclosporin, cyclophosphamide, and glococorticoids; and
 - (c) a pharmaceutically acceptable carrier.